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Spectrophotometric Determination of Sulfamethoxazole Drug by Using Azo Coupling Reaction

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Abstract

This research is based on a simple and good method for determining sulfamethoxazole in its pure form and pharmaceutical preparations. Spectrophotometric determination using the azo Sulfamethoxazole coupling reaction in an aqueous solution and several standard solutions were prepared. The optimal conditions were also studied. The effect of the type of acid and its volume, the influence of the base type and its volume, the effect of reagent volume (100 µg/mL), the effect of reaction time on color product stability and effectiveness, the solvent effect, and the effect of temperature on the formation and stability of colored products The product for the sulfamethoxazole drug an orange complex at a wavelength of 489 nm. This method follows Beer's law in one respect of concentrations ranging from 1–12 µg/ml and is the molar absorbance (39534.061 L/mol.cm). Sandal's sensitivity (0.0009 µg/cm), detection limit (0.3539 µg/ml), correlation coefficient value 0.999, and RSD% value 0.0010925, By applying the method to a pharmaceutical preparation containing sulfamethoxazole, the sulfamethoxazole recovery value is between 100.388075%.

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1. INTRODUCTION

Sulfamethoxazole (SMZ) belongs to the class of antibacterial sulfamides. Its scientific The name is 4-amino-N-(5-methyl-3-isoxazolyl)-benzenesulfonamid. It has the molecular formula C10H11N3O3S, and its molecular mass is 253.279 g/mol [1]. It is a synthetic antibiotic derived from sulfamate which acts as an antibacterial. Sulfamethzole is known by another name, which is neonomine [2, 3]. It is an almost odorless and tasteless compound [4]. It is white in color or is a yellowish-white powder [5] in solvents such as ether and chloroform, which are low in the water but melt in acetone and ethanol, which in turn melt in alkaline hydroxide solutions [6]. The combination of a fixed drug from Trimethoprim with sulfamethoxazole is more effective and has a short- to medium-range effect. It's rapidly absorbed after oral intake by85.5–90% and the maximum absorption is about 1-4 hours after taking the dose. [7] Sulfamethoxazole binds 70% of the proteins in blood plasma, mainly aluminum. Approximately 84.5% percent of the dose is released within 72 hours of treatment being administered [8, 9]. It is used to treat a wide range of infections, including middle ear infections, urinary tract infections, respiratory tract infections, and intestinal infections [10]. Among the most important side effects that result from treatment are digestive system disorders and abnormalities of blood components such as thrombocytopenia and rare agranulocytosis. In 1858, the scientist Peter Kress discovered that diazonium salts are prepared from nitrogenous ortho-aminophenol with nitrous acid [11].

Diazonium salts are generally prepared by treating aromatic amine with nitrous acid and increasing mineral acid [12]. Aromatic and fate amines can form diazonium salts and are stable. High at low temperatures, but the most stable compounds are aromatic as part of the benzene ring that possesses resonance. Nitrification is usually done at a very low temperature, below zero degrees Celsius [13]. Several researchers have developed a new and sensitive, simple, and reproducible HPLC for the determination of SMZ and TMP in blood plasma samples. The method requires the removal of the protein with perchloric acid using a reversed-phase HPLC device at a wavelength of 240 nm, which gave concentrations of 6.2–9.8%, a recovery rate of 88.5%, and an amount of quantitative detection of 0.39–50 µg mL. The method has successfully evaluated drugs in pharmaceutical preparations [14]. Developing a spectroscopic and sensitive method for the determination of sulfamethoxazole. This method included ion pairs formed between the nitrogen of the drug sulfamethoxazole and monochrome black. A product was extracted by chloroform, and an orange dye was obtained at 490 nm. The molar absorbance was (6.471 x 10^4) L mol cm2, and the correlation coefficient was (0.9998). The method has been applied to pharmaceutical preparations [15]. A rapid and sensitive spectrophotometer using the flow injection technique was also developed to estimate sulfamethoxazole, which relies on sulfonamide with sodium nitrite and then conjugates a compound with alpha-naphthyl amine. The range of linearity was within 0.2–20 mg/mL, and the limit of detection was 0.06 mg/mL. This is the way it is applied to pharmaceutical preparations [16].

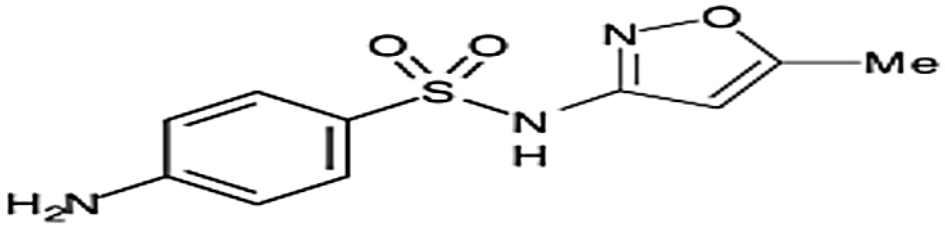


Figure. 1: Structure of Sulfamethoxazole

2. METHOD

2.1. lnstrumentel used: UV-visible spectrophotometric Shimadzu, 800, Japan. Water bath - Velp Scientific made in Europe. Center Fudge: Geemy company plc-03, Taiwan. Electrical balance - Kern&SOHN GmbH, China.

2.2. Materials: All chemicals used in this work were purchased from SDI, Merck, and BDH Companies.

2.3. Preparation of standard solution

Standard solution Sulfamethoxazole 1000 µg/ml was prepared by dissolving 0.1 gm in a Volumetric flask (100 ml) and filling the volume to the mark with distilled water [17]. The standard solution of the reagent (β-Naphthol) 1000 µg/ml was prepared by dissolving 0.1 gm in 20 ml of ethanol and then added in a volumetric flask 100 ml and completed to the mark with distilled water [18]. Sodium nitrite (1% w/v) was prepared by dissolving 1 gm in water in a volumetric flask of 100 ml and Filling the volume with distilled water up to the mark. sulfamic acid (1% w/v) was Prepared by adding 1 gm of sulfamic acid to water in a volumetric flask of 100 ml and completed to the mark with distilled water. Phosphoric acid 1 M was prepared by adding 6.22 ml of concentrated phosphoric acid 16.07 M in a volumetric flask 100 ml and filling the volume with distilled water to mark. Sulfuric acid 1M was prepared by adding 5.44 ml of concentrated sulfuric acid with a concentration of 18.38 M in a volumetric vial of 100 ml capacity and filling the volume with distilled water to the mark. Acetic acid 1 M was prepared by adding 5.74 ml of concentrated acetic acid at a concentration of 17.431 M in a volumetric flask of 100 ml and filling the volume with distilled water. Nitric acid 1 M was prepared by adding 6.97 ml of concentrated nitric acid at a concentration of 14.339 M in a volumetric flask of 100 ml and filling the volume with distilled water to the mark. Hydrochloric acid 1 M was prepared by adding 8.40 ml of hydrochloric acid at a concentration of 11.96 M in a volumetric vial of 100 ml and filling the volume with distilled water to the mark. Sodium hydroxide 1 M was prepared by dissolving 4 gm of sodium hydroxide in a volumetric flask with a capacity of 100 ml and then adding distilled water to the mark. Potassium hydroxide 1 M was prepared by dissolving 5.61 gm of potassium Hydroxide in a volumetric flask of 100 ml, and then filling the volume with distilled water. Ammonium hydroxide 1 M was prepared by dissolving 5.34 ml of concentrated ammonium hydroxide There was a concentration of 18.698 M in a volumetric flask with a capacity of 100 ml, then filling the volume Add distilled water to the mark [19].

Sodium carbonate, potassium carbonate, and sodium bicarbonate 1 M were prepared by dissolving 13.8, 10.59, and 7.2gm of these substances, respectively, in a small volume of water in a volumetric flask of 100 ml and then completing volume with distilled water to the mark [20].

2.3.1 Procedure for assay of Sulfamethoxazole in pharmaceutical preparations Tablets:
Sulfamethoxazole tablets provided by (Turkish Diva) (10), tablets were powdered and the amount of the final powder was accurately weighted to give an equivalent to about 0.1 gm of
Sulfamethoxazole was dissolved in distilled water. The prepared solution was transferred to a 100 ml volumetric flask and made up to the mark with distilled water forming a solution of 100µg.ml-1concentration.
The solution was filtered by Whitman paper to avoid suspended particles. These solutions were diluted quantitatively to form a concentration in the range of the calibration curve.

2.4. Spectrophotometric determination of Sulfamethoxazole using azo coupling reaction in aqueous solution
In the original experiments, the product of the nitrification reaction was prepared by adding (1 ml) of (100) µg/ml in a volumetric Sulfamethoxazole vial of (10 ml) capacity in an ice bath with (1 ml) of acetic acid at a concentration of (1 M) and (1 ml) From (1%) of sodium nitrite with shaking for 10 minutes, then add (1 ml) of (1%) sulfamic acid to remove the excess of sodium nitrite after five minutes of shake in ice bath add (1 ml) of (100) µg/ml of the reagent β-Naphthol directly, then followed Add (1 ml) (1 M) sodium carbonate for the coupling reaction, then left in an ice bath for 20 minutes, then we use distilled water to reduce the volume fill to the mark. To produce a colored complex was prepared that was different from the blank solution prepared through the same additives but without the drug. Several factors that affect the absorption of the resulting azo formulation were studied to obtain the best sensitivity and detection limit for determining the drug, and these conditions were studied with a wavelength according to the drug used [21]. The wavelength of the drug Sulfamethoxazole was (489) nm.

3. RESULTS AND DISCUSSION

3.1. Spectrophotometric determination of Sulfamethoxazole using azo coupling reaction in an aqueous solution
The absorption spectrum of Sulfamethoxazole the studied material was recorded using the coupling reaction of azo dyes in an aqueous solution, and a solution of concentration (100 µg/ml) was Sulfamethoxazole prepared using solvent water. Examine the drugs in these solutions by visible and UV-Vis spectroscopy in the wavelength range (200-800) nm, where the solution has Sulfamethoxazole a high absorbance of (0.394) at the wavelength (489) nm and record the wavelength as all subsequent The maximum wavelength measured by the material [22]

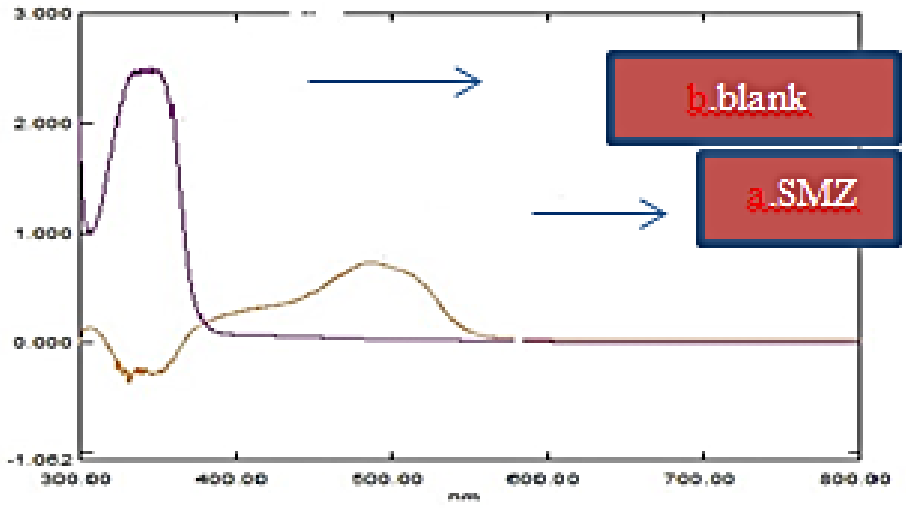


Figure 2:abosrption spectrum of(a) SMZ solution and (b) blank solution

3.2 Study the best conditions reaction:
A series of experiments to study the Effect of some factors affecting the spectral assessment of the drug used in this study.

3.2.1. Effect of the type of acid:
This study was conducted using a series of experiments, as acid solutions were prepared with A concentration is (0.5 M); the results obtained were summarized in Table (1) and Figure (3), which clearly show that the best acid is Phosphoric acid Sulfamethoxazole a concentration of (0.5 M) as it gave the highest absorbance (0.394) [23].

Table (1): Absorption data for the Effect of the acid type

1 ml From 0.5M different acids	HCl	H_2SO_4	HNO_3	H_3PO_4	CH_3COOH
absorb λmax = 489 nm	0.387	0.354	0.211	0.394	0.326

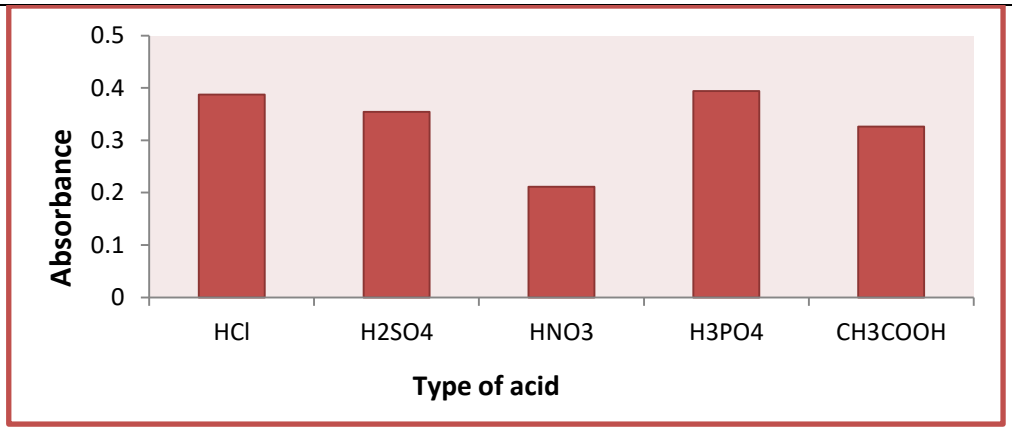


Figure (3): Effect of acid type on the absorbance of Sulfamethoxazole

3.2.2. Effect of acid volume:
This study was carried out using a series of experiments, as different volumes of phosphoric acid were prepared at concentrations of 0.5 M. The obtained results are sulfamethoxazole, as summarized in Table (2), This clearly shows the best volume of acid is 0.8 ml of phosphoric acid with a concentration of 0.5 M, as it gave the highest absorption (0.428). Figure (4) shows that with increasing the volume of the acid, the absorbance increases, and suddenly it decreases because the primary amine (the drug) becomes inactive. The ideal volume for the highest absorption was detected in subsequent experiments (0.8) ml of phosphoric acid with sulfamethoxazole [24].

Table (2) The effect of acid volume on the absorbance of the colored product of SMZ

V of 0.5M acids	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
Ab at λ max = 489	0.382	0.385	0.391	0.398	0.401	0.406	0.411	0.428	0.408	0.384

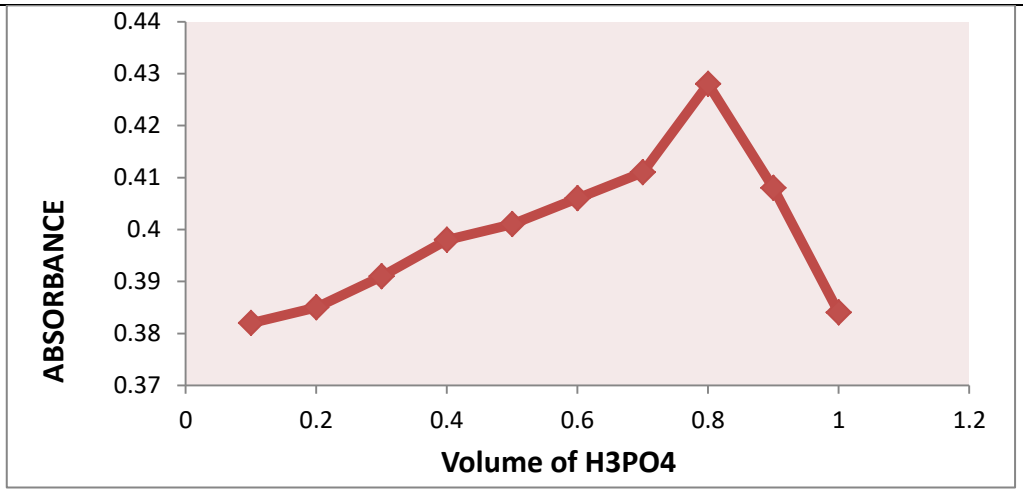


Figure (4): The effect of the volume of phosphoric acid at a concentration of 0.5 M

3.2.3. The influence of the base type:

Base solutions were prepared with concentrations of (0.5 M); Table (3) and Figure (5) clearly show that the best base for Sulfamethoxazole is Potassium hydroxide with a concentration of M (0.5) as it gave the highest absorbance (0.451) [25].

Table (3): Effect of the type of base on the absorbance of the colored products of (SMZ)

1ml from 0.5M base	NaOH	KOH	NH_4OH	Na_2CO_3	K_2CO_3	$NaHCO_3$
Ab at λ max = 489	0.428	0.451	0.366	0.275	0.383	0.388

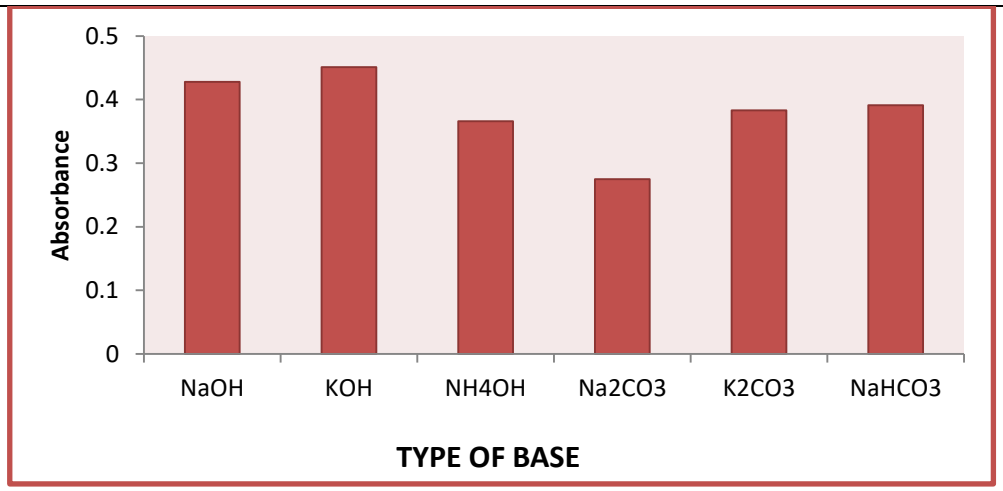


Figure (5): Effect of the base type on the absorbance of the colored product (SMZ)

3.2.4. The effect of the ideal volume of (0.5 molars) of (KOH):

Volumes of (1-10) ml of the base were prepared with concentrations of (0.5 M) of Potassium hydroxide. The results obtained were summarized in Table (4) and Figure (6), which clearly show that the best base volume for the drug Sulfamethoxazole is (0.3 ml) of the base with a concentration of Potassium hydroxide (0.5 M), as it gave the highest absorption (0.551).

Table (4): Effect of base size on the absorbance of the stained product of Sulfamethoxazole

V of 0.5M bases	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
Ab at λ max = 489	0.542	0.546	0.551	0.549	0.531	0.520	0.501	0.486	0.471	0.450

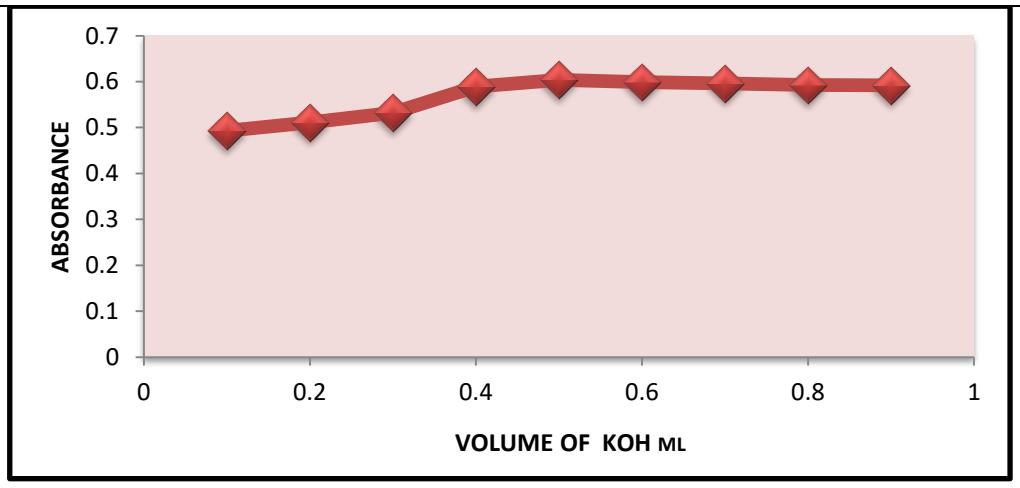


Figure (6): Effect of ideal base size (potassium hydroxide) on absorption of colored products (SMZ)

3.2.5. The effect of the optimal volume of 1% of sodium nitrite:

Variable volumes of sodium nitrite were taken from the solution, whose concentration was (1%). The results obtained are summarized as follows in Table (5), which clearly shows that the ideal volume of sodium nitrite for Sulfamethoxazole is (0.4 ml) at a concentration of (1%), as it gave the highest absorption (0.613) [26].

Table (5): Effect of 1% Volume sodium nitrite on the absorption of colored Sulfamethoxazole products

V of 1% Sodium Nitrite	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
Ab at λ max = 489	0.591	0.603	0.608	0.613	0.601	0.581	0.579	0.563	0.558	0.551

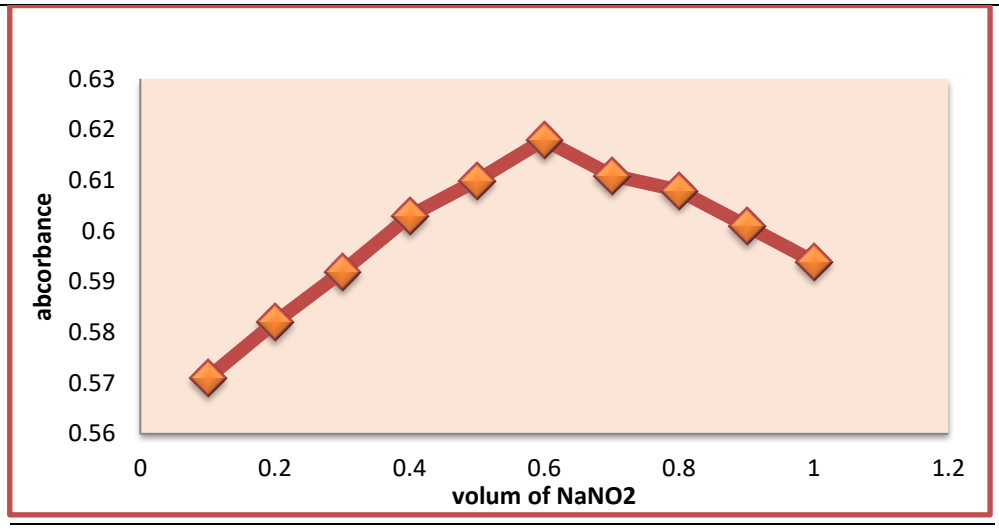


Figure (7): Effect of 1% volume sodium nitrite on the absorption of colored Sulfamethoxazole products (SMZ)

3.2.6. Perfect Size Effect 1% Sulfamic Acid.

Variable volumes of sulfamic acid were taken from the solution, whose concentration was (1%). The obtained results are summarized in Table (6), which clearly shows that the best ideal volume of sulfamic acid for Sulfamethoxazole is (0.6 ml) at a concentration of (1%), as it gave the highest absorption (0.618).

Table (6): Effect of 1% volume of sulfamic acid on Sulfamethzole

V of 1% Sulfamic acid	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
Ab at λ max = 489	0.607	0.609	0.611	0.614	0.617	0.618	0.616	0.615	0.613	0.612

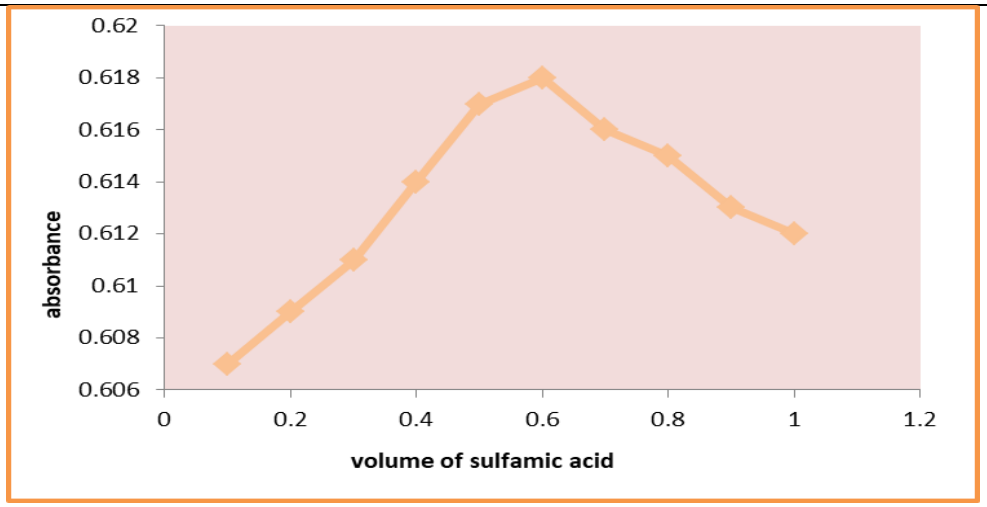


Figure (8): Effect of 1% volume of Sulfamic acid on the absorbance of the colored product (SMZ)

3.2.7. Effect of reagent volume (100 µg ml^{-1}):

Variable volumes of β-Naphthol reagent were taken at a concentration (100 µg ml^{-1}) with Sulfamethoxazole. The obtained results were summarized in Table (7) and Figures (9), which clearly show the best ideal volume of β-Naphthol reagent (0.7 ml) for Sulfamethoxazole, as it gave the highest absorbance (0.630). Absorption increases with increasing reagent volume, but absorption decreases when reagent volume is increased beyond the required limit because this volume is not suitable for conjugation with the drug [27, 28].

Table (7): The effect of the reagent volume at a concentration (100 µg ml^{-1}) on the absorbance of Sulfamethoxazole

V of (100 µg ml^{-1}) Reagent	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
Ab at λ max = 489	0.591	0.595	0.605	0.611	0.623	0.627	0.630	0.625	0.621	0.618

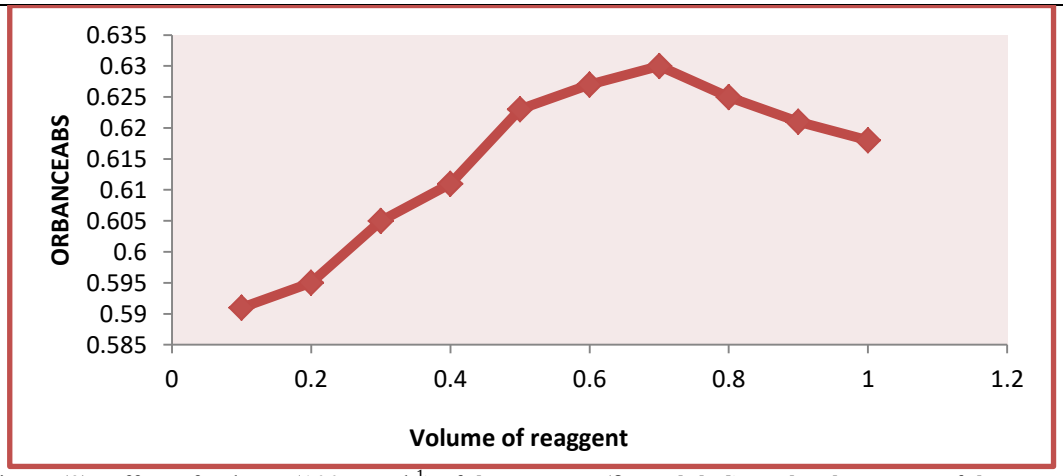


Figure (9): Effect of volume (100 µg ml^{-1}) of the Reagent (β-Naphthol) on the absorption of the stained product for SMZ

3.2.8. Effect of reaction time on product color stability:

Different times were taken for the reaction (5-65) minutes, Table (8), Figure (10). It was noted that the best time to complete the reaction was (45minutes) for Sulfamethoxazole, as it gave the highest absorbance (0.671).

Table (8): Effect of reaction time on the stability of SMZ-colored products

Time (min)	5	10	15	20	25	30	35	40	45	50	55	60	65
Ab at λ max = 489	0.570	0.582	0.598	0.606	0.631	0.638	0.645	0.669	0.671	0.656	0.643	0.531	0.520

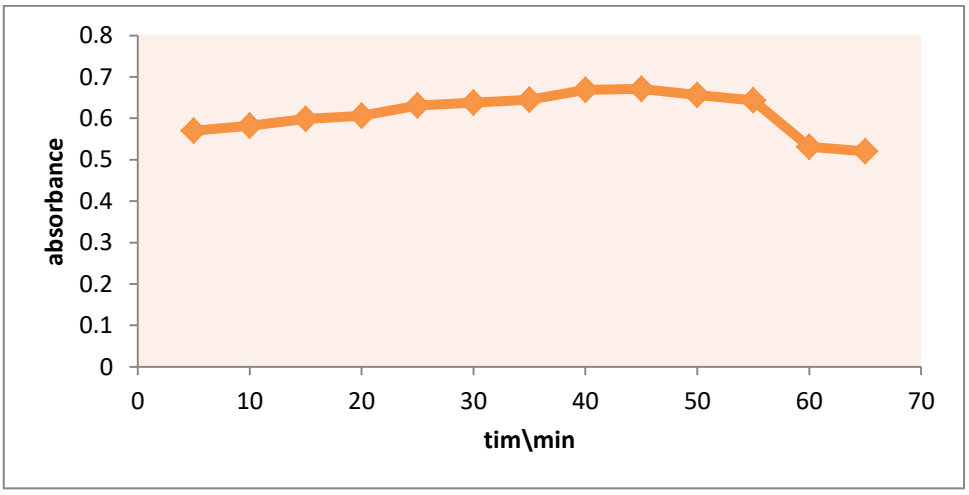


Figure (10): Effect of reaction Time on the stability of SMZ-colored products

3.2.9. Effect of adding sequence

As different sequences of additives were taken, Table (9) and Figure (11) show the results. It was observed that the best addition sequence is (1), as it gave the highest absorbance (0.670) [29].

Table (9): Shows the Effect of the addition sequence

No.	1	2	3	4	5	6
Addition	D+H+N+S+R+B	R+H+N+S+D+B	D+H+N+B+R+S	D+B+R+N+H+S	R+B+D+H+N+S	R+H+N+B+D+S
λ_{max}=489	0.670	0.451	0.012	0.254	0.019	0.226

D: (Sulfamethoxazole), H: (Phosphoric acid), S: (sulfamic acid), N: (Sodium nitrite), R: (β-Naphthol), B: (Potassium hydroxide).

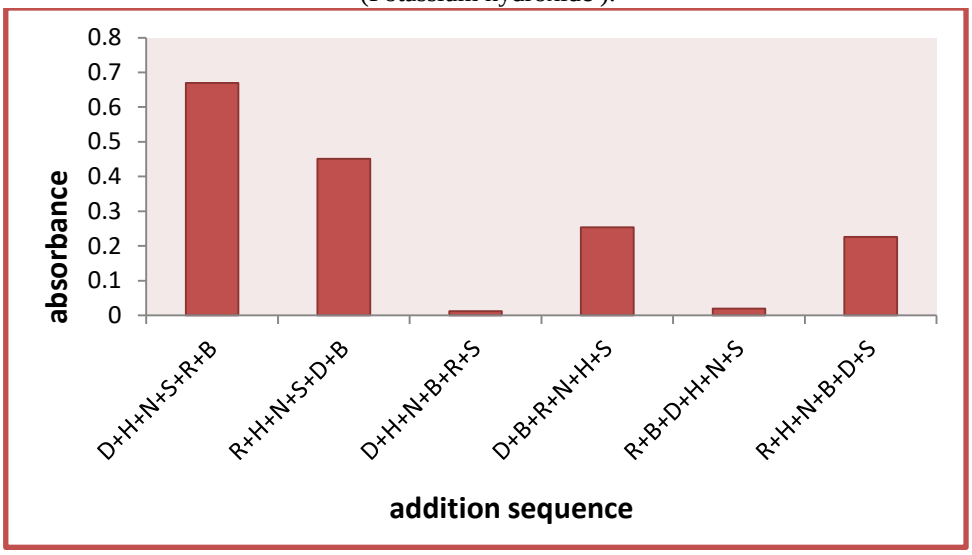


Figure (11): Effect of addition order on absorption of (SMZ) colored products

3.2.10. Solvent effect:

Different solvents (water, ethanol, methanol, acetonitrile, 1-propanol, and acetone) were taken. The results obtained are summarized as follows in Table (10) and Figures (12), This clearly shows best solvent is (water) for Sulfamethoxazole (0.671). Water solvent is considered one of the best solvents because it is easy to obtain and cheap. It is also considered one of the safe and environmentally friendly solvents [30].

Table (10) Effect of solvents on the absorption of colored product sulfamethoxazole

No.	1	2	3	4	5	6
Solvent	Water	Ethanol	Methanol	Acetonitrile	1-Propanol	Acetone
λ_{max}=489	0.671	0.653	0.638	0.439	0.589	0.641

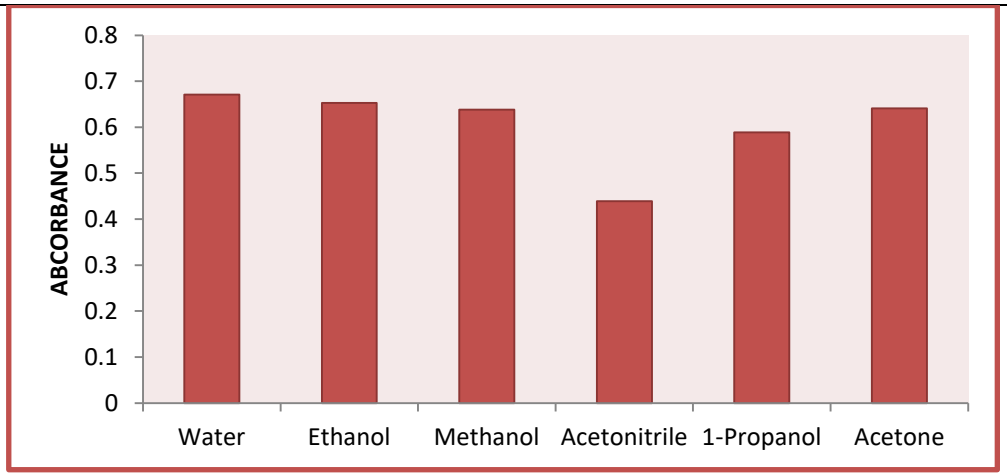


Figure (12): Effect of solvents on the absorption of colored product sulfamethoxazole drug complex

3.3. The nature of the resulting product.

3.3.1. Method of Continuous Variation (Job's Method)

The obtained results are summarized in Table (12), which clearly shows that the best volume of the drug and reagent is (0.5 ml) for sulfamethoxazole, as gave the highest absorbance (0.352) [32].

Table (12): Data of the continuous change method for Sulfamethoxazole β -Naphthol

Drug volume/ml	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9
Reagent volume/ml	0.9	0.8	0.7	0.6	0.5	0.4	0.3	0.2	0.1
Ab of SMZ	0.75	0.132	0.221	0.278	0.352	0.303	0.254	0.206	0.096

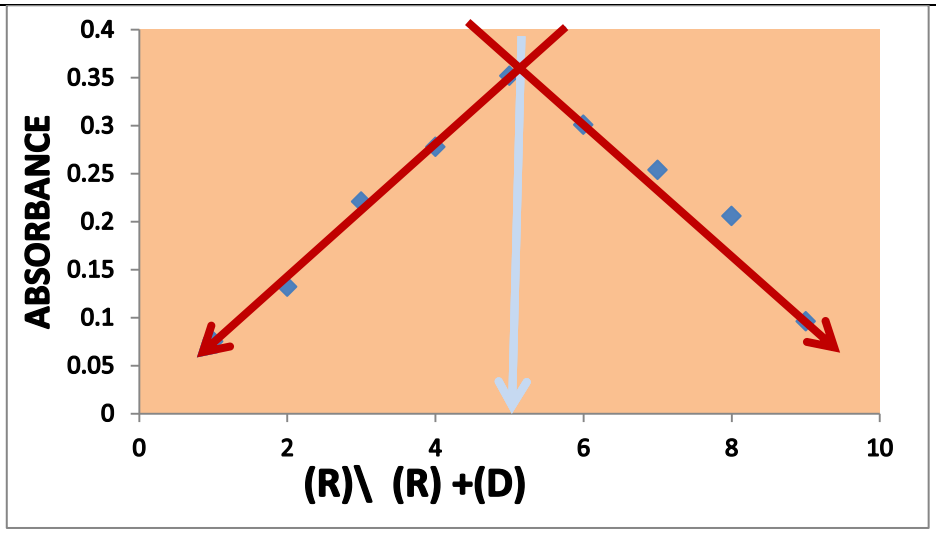


Figure (12): The method of continuous changes (JOB) for(SMZ)

3.3.2.Mole ratio:

The results are summarized in Table (13), which clearly shows that the greater the size of the reagent, the greater the absorbance of Sulfamethoxazole, as gave the highest absorbance (1.236) [33].

Table (12): Absorbance values for the results of the molar ratios method for Sulfamethoxazole: β-Naphthol

Reagent volume/ml	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5
Absorbance at λmax =489nm	0.622	0.954	1.023	1.093	1.134	1.189	1.205	1.236	1.245

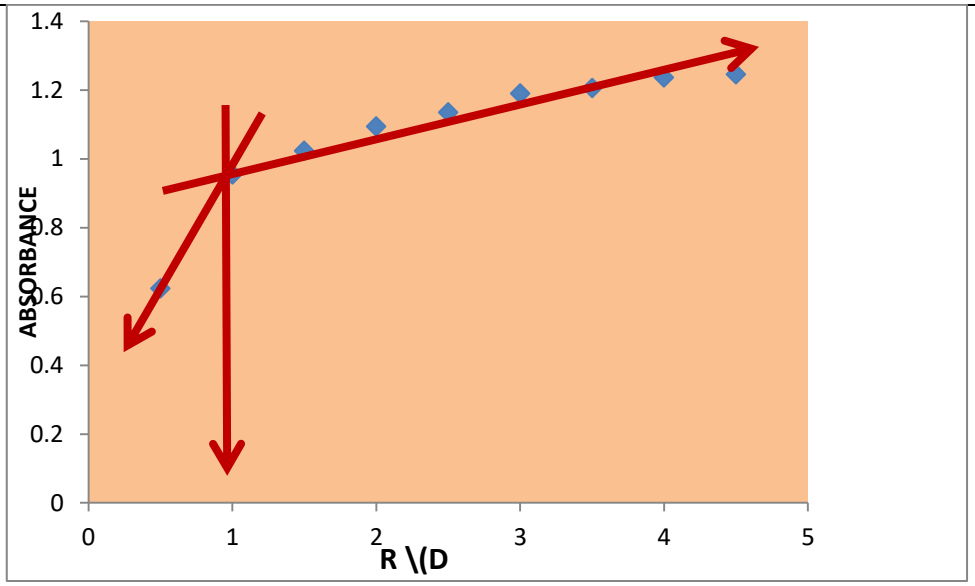
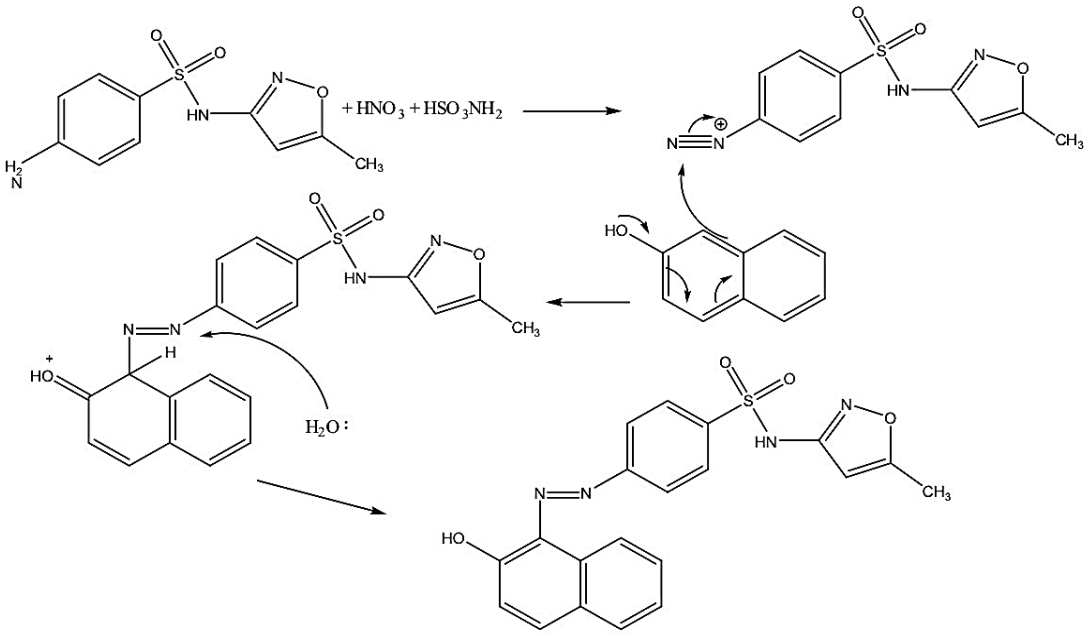


Figure (13): mole ratio method for SMZ drug



Figure(15): Proposed mechanism for the formation of the colored product of Sulfamethoxazole

3.4. Calibration curve for Sulfameth0xazole complexed with β-naphthol:

In several volumetric vials with a capacity of (10 ml), (variable volumes of (SMZ) drug ranging from (0.1-1.2) ml were taken, with concentrations (1-12 µg/ml), (0.8 ml) of Phosphoric acid, (0.4 ml) of sodium nitrite, (0.6 ml) of sulfamic acid and (0.7 ml of β-naphthol), (0.3 ml) of Potassium hydroxide, the volume is completed to the mark with distilled water, the absorbance is measured at the highest wavelength against the blank solution. Figure (16) shows the calibration curve for the drug (SMZ) with concentrations that obey Beer's law within the range of (1-12) µg ml^{-1}. The molar absorption coefficient of the product is (39534.061 L/mol.cm), and Sandal's sensitivity is (0.0009 µg/cm^2) [34, 35].

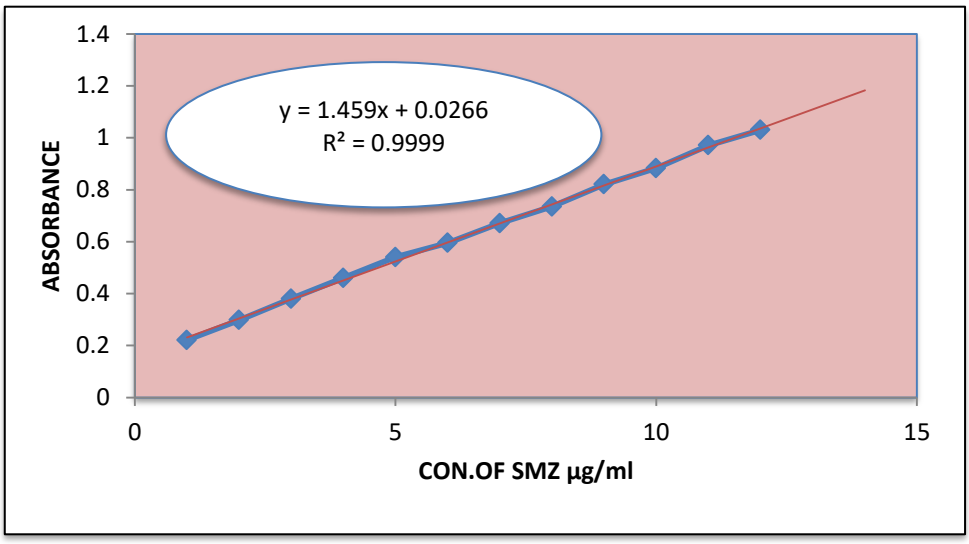


Figure (16): Calibration curve for (SMZ) drug

3.5. Interference effect:

To find out the effect of the interfering substances with the drug, each of the interfering substances (lactose, starch, Arabic Gum, glucose, Talc, Calcium phosphate)was used (1 ml) and concentration (1000 mg/ml) with (1 ml), (100mg⁄ml) of each of the two drugs. The remainder of the additive is supplemented to its ideal volumes and then diluted with distilled water in a 10ml volumetric flask. Then the absorbance is measured at (489) nm for sulfamethoxazole. The results are shown in Table (13) for Sulfamethoxazole. The results in this table show that there is no effect of the interactions on Sulfamethoxazole in pharmaceutical preparations [36, 37].

Table (13): Effect of Interactions on the Absorption of Sulfamethoxazole

No.	100ppm interference	Abs.	Recovery %	E_{rel}%
1	Lactose	0.331	96.8886	-3.111782
2	Starch	0.335	98.0966	-1.903323
3	Arabic Gum	0.337	98.7009	-1.299094
4	Glucose	0.336	98.3987	-1.601208
5	Talc	0.328	95.9818	-4.018127
6	$Ca_3(PO_4)_2$	0.319	93.2628	-6.73716

3.6. Drug detection limits and quantity limits

The detection and quantitative detection limits were calculated by taking ten iterations of the blank solution [38]. As shown in Table (14).

Table (14): Calculating the detection limit and the quantitative limit for Sulfamethoxazole

Parameters	$\bar{x}_B$	$S_B = [(X_i-\bar{x})^2/n-1]^{1/2}$	OD = $\bar{x}_B$+3 S_B	LOQ = $\bar{x}_B$ + 10 S_B
Sulfamethoxazole	0.019	0.1140	0.3539	0.1259

3.7. Colored output stability constant.

Depending on my method, molar ratios, and previous continuous changes, the ratio of [drug: reagent] is [1:1] and the stability constant of the complex. The value of High stability constant. Hence, the dye formed is very stable, as shown in Table (15).

Table (15): Stability constant data for the colored product of (SMZ)

V $4x10^{-4}$M of SMZ /ml	Final con. SMZ/M	As*	Am*	α	K(L..Mol^{-1})	Mean of K (L.Mol^{-1})
0.3	$1.2x10^{-5}$	0.453	0.459	0.0342	2487.1209	
0.5	$2x10^{-5}$	0.758	0.763	0.2304	6978.8358	23958.3845
0.7	$2.8x10^{-5}$	1	1.04	1.972	2301.88	

3.8. Accuracy and precision testing:

Sulfamethoxazole's accuracy and precision test were calculated using four concentrations of the calibration curves (12, 9, 6, 3). Table (16) shows the effect of accuracy and precision. Five replicates were taken, and the optimal conditions of the method were applied. The results of this method are of good accuracy and precision, by the value of the recovery rate of (100.3880%).

Table (16): Accuracy and precision data for the proposed method for the determination of Sulfamethoxazole

Amount of SMZ /µg mL^{-1}	*Found	Recovery %	Average Recovery %	E_{rel}%	Average E_{rel}%	RSD%
12	12.0288	100.24	100.388075	0.2400	0.388325	0.00117
9	9.0144	100.16		0.1600		0.0002
6	6.0253	100.4223		0.4223		0.0006
3	3.0219	100.73		0.73		0.0024

3.9. Applications of the proposed method to Medicines:

pharmaceutical preparations were used (Turkish Diva Company), which contain (10 mg) of Sulfamethoxazole every 1gm, and the sample is prepared. The results shown in Table (17) are confirmed. The proposed method's success in determining Sulfamethoxazole in the used preparation.

Table (17): Data for the determination of Sulfamethoxazole in the pharmaceutical preparation (Bactrim)

Amount of SMZ /µg mL^{-1}	*Found	Recovery %	Average Recovery %	E_{rel}%	Average E_{rel}%	RSD%
12	11.754	97.95	96.46195	-2.05	-3.44805	0.0014
9	8.7436	97.1511		-2.4889		0.0002
6	5.7708	96.18		-3.82		0.0006
3	2.9276	97.5867		-2.4133		0.0128

Table (18): Statistical results of the proposed spectral method for drug estimation (SMZ)

Parameter	**Sulfamethoxazole & β-Naphthol**
Product Color	Orange
λ max	489nm
regression equation	y = 1.459x + 0.0266
The standard deviation of the regression	0.26415
Correlation coefficient (r)	0.999
C.L for slop ($b \pm tS_b$) at 99%	1.459 ± 0.136234
C.L for Intercept ($b \pm tS_a$) at 99%	0.0266 ± 0.615457
Concentration range (µg ml^{-1})	(1-12) µg ml^{-1}
Limit of Detection (µg ml^{-1})	0.3539
Limit of Quantitative (µg ml^{-1})	0.1259
Sandals Sensitivity (µg ml^{-1})	0.0009
Molar absorbance (L/mol.cm	39534.061
Product ingredients	1:1
recover %	100.38807
RSD% n=5	0.0010925
C.L.Con.12 (µg ml-1)	12.088±0.04093
C.L. Con.9 (µg ml-1)	9.0144 ±0.00374
C.L con.6(µg ml^{-1})	6.0253 ±0.00693
C.L con.3(µg ml^{-1})	3.0219±0.01503

4. CONCLUSION

In this research, a new analytical method was developed for determining Sulfamethoxazole in the pharmaceutical preparation using the azo coupling reaction, we are encouraging, a new and effective reagent used to estimate the drug and study the optimal conditions. This method gave good and fast results, and it was an economical, more sensitive, and selective method in drug estimation. The possibility of using this method to estimate drugs in the pure state and their pharmaceutical preparations and use them in very small quantities

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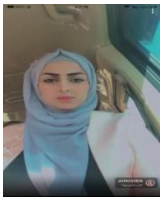

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